



1

SEQUENCE LISTING

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BRANDT, SILKE
RIEKE, ERWIN
SOBEL, CORNELIUS
LO, KIN-MING
WAY, JEFFREY C.
GILLIES, STEPHEN

<120> ERYTHROPOIETIN FORMS WITH IMPROVED PROPERTIES

<130> MERCK-2056

<140> 09/708,506

<141> 2000-11-09

<150> 60/164,855

<151> 1999-11-12

<160> 26

<170> PatentIn Ver. 2.1

<210> 1

<211> 514

<212> DNA

<213> Homo sapiens

<220>

<221> CDS

<222> (8)..(505)

<223> Human EPO, DNA sequence modified but no change in
protein sequence

<400> 1

cccgggg gcc cca cca cgc ctc atc tgt gac agc cga gtg ctg gag agg 49
Ala Pro Pro Arg Leu Ile Cys Asp Ser Arg Val Leu Glu Arg
1 5 10

tac ctc ttg gag gcc aag gag gcc gag aat atc acg acc ggc tgt gct 97
Tyr Leu Leu Glu Ala Lys Glu Ala Glu Asn Ile Thr Thr Gly Cys Ala
15 20 25 30

gaa cac tgc agc ttg aat gag aac atc acc gtg cct gac acc aaa gtg 145
Glu His Cys Ser Leu Asn Glu Asn Ile Thr Val Pro Asp Thr Lys Val
35 40 45

aat ttc tat gcc tgg aag agg atg gag gtt ggc cag cag gcc gta gaa 193
Asn Phe Tyr Ala Trp Lys Arg Met Glu Val Gly Gln Gln Ala Val Glu
50 55 60

gtg tgg cag ggc ctg gcc ctg ctg tcg gaa gct gtc ctg cgg ggc cag 241
Val Trp Gln Gly Leu Ala Leu Ser Glu Ala Val Leu Arg Gly Gln
65 70 75

gcc ctg ttg gtc aac tct tcc cag ccg tgg gag ccc ctg caa ctg cat 289
Ala Leu Leu Val Asn Ser Ser Gln Pro Trp Glu Pro Leu Gln Leu His
80 85 90

gtg gat aaa gcc gtg agt ggc ctt cgc agc ctc acc act ctg ctt cgg 337
Val Asp Lys Ala Val Ser Gly Leu Arg Ser Leu Thr Thr Leu Leu Arg
95 100 105 110

gct ctg gga gcc cag aag gaa gcc atc tcc cct cca gat gcg gcc tca 385
Ala Leu Gly Ala Gln Lys Glu Ala Ile Ser Pro Pro Asp Ala Ala Ser
115 120 125

gct gct ccc ctc cgc aca atc act gct gac act ttc cgc aaa ctc ttc 433
Ala Ala Pro Leu Arg Thr Ile Thr Ala Asp Thr Phe Arg Lys Leu Phe
130 135 140

cga gtc tac tcc aat ttc ctc cgg gga aag ctg aag ctg tac aca ggg 481
Arg Val Tyr Ser Asn Phe Leu Arg Gly Lys Leu Lys Leu Tyr Thr Gly
145 150 155

gag gcc tgc cgg aca ggg gat aga tgactcgag 514
Glu Ala Cys Arg Thr Gly Asp Arg
160 165

<210> 2
<211> 166
<212> PRT
<213> Homo sapiens

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Ala Pro Pro Arg Leu Ile Cys Asp Ser Arg Val Leu Glu Arg Tyr Leu
1 5 10 15

Leu Glu Ala Lys Glu Ala Glu Asn Ile Thr Thr Gly Cys Ala Glu His
20 25 30

Cys Ser Leu Asn Glu Asn Ile Thr Val Pro Asp Thr Lys Val Asn Phe
35 40 45

Tyr Ala Trp Lys Arg Met Glu Val Gly Gln Gln Ala Val Glu Val Trp
50 55 60

Gln Gly Leu Ala Leu Leu Ser Glu Ala Val Leu Arg Gly Gln Ala Leu
65 70 75 80

Leu Val Asn Ser Ser Gln Pro Trp Glu Pro Leu Gln Leu His Val Asp
85 90 95

Lys Ala Val Ser Gly Leu Arg Ser Leu Thr Thr Leu Leu Arg Ala Leu
100 105 110

Gly Ala Gln Lys Glu Ala Ile Ser Pro Pro Asp Ala Ala Ser Ala Ala
115 120 125

Pro Leu Arg Thr Ile Thr Ala Asp Thr Phe Arg Lys Leu Phe Arg Val
130 135 140

*See
D1
Cont*

Tyr Ser Asn Phe Leu Arg Gly Lys Leu Lys Leu Tyr Thr Gly Glu Ala
 145 150 155 160

Cys Arg Thr Gly Asp Arg
 165

<210> 3
 <211> 52
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: Oligo1

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<210> 4
 <211> 49
 <212> DNA
 <213> Artificial Sequence

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 <223> Description of Artificial Sequence: Oligo2

<400> 4
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<210> 5
 <211> 52
 <212> DNA
 <213> Artificial Sequence

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 <223> Description of Artificial Sequence: Oligo3

<400> 5
 ctgcagcttg aatgagaaca tcaccgtgcc tgacaccaaa gtgaatttct at 52

<210> 6
 <211> 48
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: Oligo4

<400> 6
 gcctggaaga ggatggaggt tggccagcag gccgtagaag tgtggcag 48

<210> 7
 <211> 51

*Sub
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 Cont*

<212> DNA
 <213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Oligo5

<400> 7

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<210> 8

<211> 49

<212> DNA

<213> Artificial Sequence

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<223> Description of Artificial Sequence: Oligo6

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aactcttccc agccttggga gcccctgcaa ctgcatgtgg ataaagccg 49

<210> 9

<211> 52

<212> DNA

<213> Artificial Sequence

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<223> Description of Artificial Sequence: Oligo7

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<210> 10

<211> 48

<212> DNA

<213> Artificial Sequence

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<223> Description of Artificial Sequence: Oligo8

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ggaagccatc tccctccag atgcggcctc agctgtccc ctccgcac 48

<210> 11

<211> 53

<212> DNA

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<211> 59
<212> DNA
<213> Artificial Sequence

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<210> 13
<211> 49
<212> DNA
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<400> 13
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<210> 14
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<212> DNA
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<223> Description of Artificial Sequence: Oligo12

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<210> 15
<211> 49
<212> DNA
<213> Artificial Sequence

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<223> Description of Artificial Sequence: Oligo13

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<210> 16
<211> 48
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Oligo14

<400> 16
ggaagccatc tcccctccag atgcggccgc agctgtccc ctccgcac 48

Full
D.I.
Cont

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 <211> 232
 <212> PRT
 <213> Homo sapiens

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 <223> Human IgG1 Fc region-mature protein

<400> 17

Glu Pro Lys Ser Cys Asp Lys Thr His Thr Cys Pro Pro Cys Pro Ala
 1 5 10 15

Pro Glu Leu Leu Gly Gly Pro Ser Val Phe Leu Phe Pro Pro Lys Pro
 20 25 30

Lys Asp Thr Leu Met Ile Ser Arg Thr Pro Glu Val Thr Cys Val Val
 35 40 45

Val Asp Val Ser His Glu Asp Pro Glu Val Lys Phe Asn Trp Tyr Val
 50 55 60

Asp Gly Val Glu Val His Asn Ala Lys Thr Lys Pro Arg Glu Glu Gln
 65 70 75 80

Tyr Asn Ser Thr Tyr Arg Val Val Ser Val Leu Thr Val Leu His Gln
 85 90 95

Asp Trp Leu Asn Gly Lys Glu Tyr Lys Cys Lys Val Ser Asn Lys Ala
 100 105 110

Leu Pro Ala Pro Ile Glu Lys Thr Ile Ser Lys Ala Lys Gly Gln Pro
 115 120 125

Arg Glu Pro Gln Val Tyr Thr Leu Pro Pro Ser Arg Glu Glu Met Thr
 130 135 140

Lys Asn Gln Val Ser Leu Thr Cys Leu Val Lys Gly Phe Tyr Pro Ser
 145 150 155 160

Asp Ile Ala Val Glu Trp Glu Ser Asn Gly Gln Pro Glu Asn Asn Tyr
 165 170 175

Lys Thr Thr Pro Pro Val Leu Asp Ser Asp Gly Ser Phe Phe Leu Tyr
 180 185 190

Ser Lys Leu Thr Val Asp Lys Ser Arg Trp Gln Gln Gly Asn Val Phe
 195 200 205

Ser Cys Ser Val Met His Glu Ala Leu His Asn His Tyr Thr Gln Lys
 210 215 220

Ser Leu Ser Leu Ser Pro Gly Lys
 225 230

*See
 DI
 cont*

<210> 18
 <211> 326
 <212> PRT
 <213> Homo sapiens

<220>
 <223> Human IgG2 constant region (CH1, hinge, CH2,
 CH3) - mature protein

<400> 18

Ala Ser Thr Lys Gly Pro Ser Val Phe Pro Leu Ala Pro Cys Ser Arg
 1 5 10 15

Ser Thr Ser Glu Ser Thr Ala Ala Leu Gly Cys Leu Val Lys Asp Tyr
 20 25 30

Phe Pro Glu Pro Val Thr Val Ser Trp Asn Ser Gly Ala Leu Thr Ser
 35 40 45

Gly Val His Thr Phe Pro Ala Val Leu Gln Ser Ser Gly Leu Tyr Ser
 50 55 60

Leu Ser Ser Val Val Thr Val Pro Ser Ser Asn Phe Gly Thr Gln Thr
 65 70 75 80

Tyr Thr Cys Asn Val Asp His Lys Pro Ser Asn Thr Lys Val Asp Lys
 85 90 95

Thr Val Glu Arg Lys Cys Cys Val Glu Cys Pro Pro Cys Pro Ala Pro
 100 105 110

Pro Val Ala Gly Pro Ser Val Phe Leu Phe Pro Pro Lys Pro Lys Asp
 115 120 125

Thr Leu Met Ile Ser Arg Thr Pro Glu Val Thr Cys Val Val Val Asp
 130 135 140

Val Ser His Glu Asp Pro Glu Val Gln Phe Asn Trp Tyr Val Asp Gly
 145 150 155 160

Val Glu Val His Asn Ala Lys Thr Lys Pro Arg Glu Glu Gln Phe Asn
 165 170 175

Ser Thr Phe Arg Val Val Ser Val Leu Thr Val Val His Gln Asp Trp
 180 185 190

Leu Asn Gly Lys Glu Tyr Lys Cys Lys Val Ser Asn Lys Gly Leu Pro
 195 200 205

Ala Pro Ile Glu Lys Thr Ile Ser Lys Thr Lys Gly Gln Pro Arg Glu
 210 215 220

Pro Gln Val Tyr Thr Leu Pro Pro Ser Arg Glu Glu Met Thr Lys Asn
 225 230 235 240

Gln Val Ser Leu Thr Cys Leu Val Lys Gly Phe Tyr Pro Ser Asp Ile
 245 250 255

*Sub
 DI
 100*

Ala Val Glu Trp Glu Ser Asn Gly Gln Pro Glu Asn Asn Tyr Lys Thr
260 265 270

Thr Pro Pro Met Leu Asp Ser Asp Gly Ser Phe Phe Leu Tyr Ser Lys
275 280 285

Leu Thr Val Asp Lys Ser Arg Trp Gln Gln Gly Asn Val Phe Ser Cys
290 295 300

Ser Val Met His Glu Ala Leu His Asn His Tyr Thr Gln Lys Ser Leu
305 310 315 320

Ser Leu Ser Pro Gly Lys
325

<210> 19

<211> 4

<212> PRT

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<223> Description of Artificial Sequence: Linker

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Ala Ala Ala Ala
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<210> 20

<211> 5

<212> PRT

<213> Artificial Sequence

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<223> Description of Artificial Sequence: Linker

<400> 20

Ala Ala Ala Ala Ala
1 5

<210> 21

<211> 4

<212> PRT

<213> Artificial Sequence

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<223> Description of Artificial Sequence: Linker

<400> 21

Gly Gly Gly Gly
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<210> 22

<211> 5

<212> PRT

*See
D1
cont*

<213> Artificial Sequence

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<223> Description of Artificial Sequence: Linker

<400> 22

Gly Gly Gly Gly Gly
1 5

<210> 23

<211> 7

<212> PRT

<213> Artificial Sequence

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<223> Description of Artificial Sequence: Linker

<400> 23

Gly Gly Gly Gly Gly Gly Gly
1 5

<210> 24

<211> 5

<212> PRT

<213> Artificial Sequence

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<223> Description of Artificial Sequence: Linker

<400> 24

Gly Gly Pro Gly Gly
1 5

<210> 25

<211> 5

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Linker

<400> 25

Gly Gly Gly Gly Ser
1 5

<210> 26

<211> 25

<212> PRT

<213> Artificial Sequence

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<223> Description of Artificial Sequence: Linker

*See
DI
cont*

<220>

<223> This linker sequence may encompass five to
twenty five amino acids is groups of (GGGGS)

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Gly Gly Gly Gly Ser Gly Gly Gly Gly Ser Gly Gly Gly Gly Ser Gly
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Gly Gly Gly Ser Gly Gly Gly Gly Ser
20 25

See
D1
Appendix